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(54) **METHOD OF DIAGNOSING OF EXPOSURE TO TOXIC AGENTS BY MEASURING DISTINCT PATTERN IN THE LEVELS OF EXPRESSION OF SPECIFIC GENES**

(75) Inventors: **Rina Das**, Rockville, MD (US); **Marti Jett**, Washington, DC (US); **Chanaka Mendis**, Falls Church, VA (US)

(73) Assignee: **The United States of America as represented by the Secretary of the Army**, Washington, DC (US)

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(58) **Field of Search** 435/6, 7.1, 325, 435/375, 91.2, 91.5; 436/536; 514/44; 536/24.31

(56) **References Cited**

FOREIGN PATENT DOCUMENTS

WO/94/26307	11/1994	(WO)	A61K/39/295
WO/97/02756	4/1997	(WO)	C12Q/1/68

OTHER PUBLICATIONS

Mendis, "Identification of alterations in gene expression in response to staphylococus enterotoxin B (SEB) using differential display (DD)", Molecular Biology of the Cell, vol. 9, No. Sup, 1998, p. 450A, Abstract.

Pomerantsev, "Expression of cereolysine AB genes in Bacillus anthracis vaccine strain ensures protection against experimental hemolytic anthrax infection", Vaccine, GB, Butterworth Scientific., Guildford, vol. 15, No. 17–18, Dec. 1, 1997, pp. 1846–1850, abstract.

Ledakis et al. "limitations of Differential Display", Biochemical and Biophysical Research Communications vol. 251, pp. 653–656, Oct. 1998.*

* cited by examiner

Primary Examiner—Robert A. Schwartzman
Assistant Examiner—Karen A Lacourciere
(74) *Attorney, Agent, or Firm*—Elizabeth Arwine

(57) **ABSTRACT**

A method of diagnosing exposure to a toxic agent comprising the steps of detecting the amount of protein/gene expression present in a sample of mammalian tissue or mammalian body fluids that has not been exposed to a toxic agent. Then the amount of protein/gene expression present in a sample of mammalian tissue or mammalian body fluids that has been exposed to the toxic agent is detected. A determination of the difference in the detected amount of protein/gene expression between the exposed and unexposed samples is made. A comparison of the difference to a library of expected protein/gene expression for predetermined toxic agents is made. Finally, an evaluation is made whether the difference indicates the exposure to a particular toxic agent. A treatment method for administering a therapeutic agent which inhibits the mechanistic pathways necessary to maintain the progression of lethal shock is also disclosed.

20 Claims, 27 Drawing Sheets